Determination of Forage Chemical Composition Using Remote Sensing

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Abstract

Traditional forage nutrient analysis from bench-top near-infrared spectroscopy (NIRS) or common laboratory chemical procedures provides accurate, point-based information, but often does not provide it in a timely way to allow changes in forage or animal management. The objective of this study is to determine the feasibility of estimating concentrations of nitrogen, neutral detergent fiber (NDF), and acid detergent fiber (ADF) of live, standing forages using a hand-held hyperspectral spectroradiometer (radiometer), and to compare these estimates to values determined via NIRS and laboratory chemical methods. Calibration equations were developed from canopy reflectance measurements from monocultures of Bermuda grass and then applied to a test data set to predict N, NDF, and ADF. Statistical analyses showed that forage composition estimates from the radiometer were equivalent to those from the NIRS. Such a remote-sensing approach would enable real-time assessment of forage quality, would allow mapping of the nutritional landscape, could be used as a tool to better manage pastures and supplements, and would assist in making harvesting decisions.

Resumen

El análisis tradicional de los nutrientes de los forrajes por espectroscopia de infrarrojo cercano (NIRS) o por los procedimientos comunes de análisis de laboratorio proveen información certera como punto de partida, pero a menudo esta información no esta a tiempo para permitir cambios en el manejo del forraje o el animal. El objetivo de este estudio es determinar la factibilidad de estimar las concentraciones de nitrógeno (N), fibra neutro detergente (FND) y fibra ácido detergente (FAD) de forrajes vivos en pie utilizando un espectroradiometro (radiómetro) hiperespectral manual y comparar estas estimaciones con los valores obtenidos vía NIRS y análisis de laboratorio, Las ecuaciones de calibración se desarrollaron a partir de medidas de la reflectancia de la copa de monocultivos de zacate Bermuda y después se aplicaron a un juego de datos de prueba para predecir N, FND y FAD. Los análisis estadísticos mostraron que las estimaciones de la composición del forraje obtenidas con el radiómetro fueron equivalentes a las obtenidas con el NIRS. Esta método de sensores remotos pudiera permitir evaluaciones a tiempo real de la calidad del forraje, permitiría mapear el paisaje nutricional y pudiera ser usado como una herramienta para un mejor manejo de los potreros y suplementos y asistiría en tomar decisiones de cosecha.

Key Words: hyperspectral radiometer, NIRS, nitrogen, neutral detergent fiber, acid detergent fiber

Introduction

Laboratory assessments of feed and forage quality date back more than 100 years to the proximate analysis system (Kellems and Church 1998; Coleman et al 1999), and more recently to neutral detergent fiber (NDF) and acid detergent fiber (ADF) techniques (Van Soest and Marcus 1964; Van Soest et al 1966; Van Soest and Wine 1968; Van Soest et al 1991). These more recent laboratory chemical procedures are the accepted standards (AOAC 1996) for estimating the nutritive potential of forages, but the results are generally point-based; some of the procedures can produce hazardous laboratory wastes, and the resulting information is often not made available in a timely fashion to effect changes in feed or livestock management due to the time needed to collect, prepare, and analyze the samples.

Beginning in the mid 1970s, bench-top near-infrared reflectance spectroscopy (NIRS) was evaluated for its potential to provide timely forage quality data comparable to the more traditional chemical procedures (Norris et al 1976). Since then, a large number of studies have been published on the use of NIRS to measure lignin, cell wall carbohydrates, starch, crude protein, dry matter digestibility, and intake among other forage quality variables (eg, Barton and Burdick 1981; Coleman et al 1995; Atanasova et al 1996).

Conventional NIRS evaluation of forage quality necessitates that calibration equations for the forage(s) under consideration be developed (Hruschka 1987). To this end, forage samples are collected from the field, dried, ground to small particle size, and scanned using a bench-top NIRS spectrophotometer. Statistical procedures are then used to develop and quantify relations between the NIR reflectance spectra and forage quality measurements that are determined by chemical procedures.

Determination of standing forage quality via field-based spectroradiometers would further reduce laborious field sampling, and would permit mapping of the nutritional landscape

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Table 1. Number of samples collected, nitrogen fertilizer treatments, hay cutting date, and precipitation received for the Bermuda grass pastures at El Reno, Oklahoma, during 1999, 2000, and 2001.

Year	n	Fertilizer treatment	Hay cut date	Rainfall				
				April	May	June	July	August
		kg/ha		mm				
1999	51	84, 168, 336	June 28 (DOY[1] 179)	125 (+45)[2]	109 (−23)	126 (+22)	107 (+42)	37 (−32)
2000	6	42, 84, 168	July 1 (DOY 183)	117 (+37)	73 (−59)	179 (+75)	69 (+4)	9 (−60)
2001	50	52, 103, 206	June 25 (DOY 176)	11 (−69)	181 (+49)	28 (−76)	24 (−41)	86 (+17)

[1]DOY indicates date of year.
[2]Number in parentheses denotes millimeters of rainfall above or below the 1960–1991, April–August normals.

in near-real time, thereby providing an extra level of information to the farm, ranch, or livestock manager. Although a number of studies have been reported successfully relating canopy-level biochemical properties of nonforage species to remotely sensed data by both hand-held and airborne platforms (eg, Chang and Collins 1983; Wessman et al 1988; Peterson et al 1988; Rock et al 1994; Johnson et al 1994; Yoder and Pettigrew-Crosby 1995; Adams et al 2000), similar studies on forages are limited. Richardson et al (1983) used a hand-held radiometer to estimate N content of Alicia Bermuda grass, and concluded that remote sensing could be a valuable tool for grazing land management. Selman (1998) collected hand-held spectroradiometer data from mixtures of C3 and C4 grasses. Using standard NIRS approaches, Selman (1998) developed calibration equations relating the field reflectance spectra to crude protein (CP), NDF, and ADF. Regression analysis of radiometer-predicted values of CP, NDF, and ADF to laboratory reference values yielded moderate r^2s of 0.66, 0.54, and 0.42, respectively, somewhat lower than that normally observed with bench-top NIRS instruments. These moderate r^2s may be due to insufficient variation expressed within the data set, because it consisted of only one year's data, or to the mixtures of C3 and C4 grasses used to develop the calibration equations.

Our objective was to further evaluate whether spectral reflectance data obtained in situ using field remote sensing instruments could be used to predict N, NDF, and ADF. More specifically, we wanted to determine if the r^2s could be improved, over that of Selman (1998), by collecting reflectance data from monocultures of a warm-season grass over multiple growing seasons. Bermuda grass was chosen for this study since it is an important forage species used to support the livestock industry over much of the southern Great Plains, south, and southeastern portion of the United States.

Materials and Methods

Study Site

The experiment was conducted from April through August of 1999, 2000, and 2001 on 4 pastures (1.6 ha each) containing monocultures of Midland, Midland 99, Worldfeeder, and Ozarka Bermuda grass (*Cynodon dacytlon* (L.) Pers.). These pastures were located at the Grazinglands Research Laboratory, El Reno, Oklahoma, and were established in 1991 on a Brewer silty clay loam (fine-loamy, mixed, thermic Udic Rhodustalfs). These pastures were managed as part of a different study, but were divided into 6 equal-sized strips receiving 1 of 3 randomly assigned fertilizer treatments (Table 1). Fertility treatments were assumed to foster different growth rates, plant vigor, and nutrient value of the plants, thus providing an ideal experimental site for the project. The pastures were cut for hay once each year during late June or early July (Table 1).

Rainfall was quite variable among the 3 sampling periods compared to the 30-year normals computed for the months of April–August. Rainfall in 1999 was about 10% above average, near average in 2000, and about 27% below average in 2001 (Table 1). Equipment malfunctions and unfavorable meteorological conditions during the 2000 sampling period limited the number of samples that were collected (Table 1).

Field Sampling

Remotely sensed data and vegetation samples were collected at random times during the early, middle, and late portions of the 1999 sampling period. As noted above, equipment failures during the 2000 sampling period limited the number of remotely sensed measurements that were made, resulting in usable data from only 1 sampling date during this time. Samples were collected weekly in 2001 between day of year (DOY) 115 and 176. The forage was cut for hay on DOY 176 and was not sampled again until DOY 226 and 233.

An SE-590 (Spectron Engineering, Denver, CO) spectroradiometer (hereafter referred to as radiometer) was used to measure solar radiation reflected from the Bermuda grass canopies in 252 wave bands covering the 368–1 100-nm region of the electromagnetic spectrum. The radiometer was mounted on a boom about 2 m above the soil surface, and had a 6° field-of-view, producing a view area with a 21-cm diameter. A single canopy measurement consisted of the average of 3 reflectance measurements made with the radiometer field of view positioned at 90° to the canopy. The center of each view area was offset from the adjacent one by about 20 cm. Before each canopy measurement, a scan was taken of a reference panel (Labsphere, North Sutton, NH) having a 99% spectral reflectance factor (over the spectral range of the radiometer). Each canopy measurement was divided by the reference panel reading to produce a reflectance factor (RF), which normalizes the data for variations in incident solar radiation during the sampling period. The RF data were then converted into an "absorbance" spectrum using

$$\text{absorbance spectrum} = \log_{10}(1/\text{RF}), \qquad [\mathbf{1}]$$

a mathematical transformation routinely used in development of calibration equations on NIRS systems (Hruschka 1987).

A 0.5 m^2 vegetation sampling frame was placed around the area viewed by the radiometer, and all vegetation within the

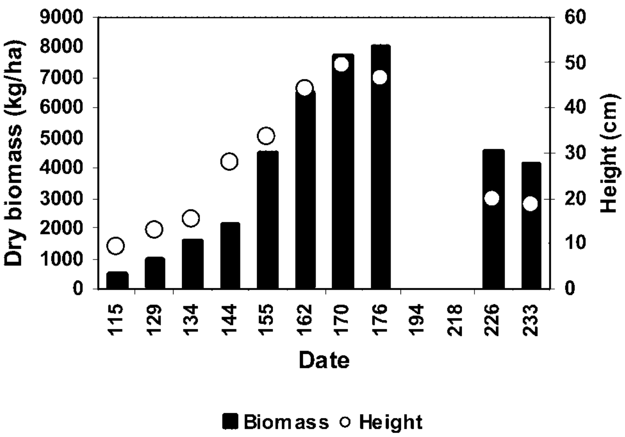

Figure 1. Average weekly biomass (dry matter basis) and height of forage observed during the 2001 study period. Average values from 1999 and 2000 fell within the range observed for 2001.

frame was clipped to ground level, bagged, and dried for 48 hours at 65°C in a forced-air oven. About one-half of the dried sample was then ground to pass a 2-mm screen using a Wiley laboratory mill, and then ground through a 1-mm screen in a cyclone mill (Udy Corporation, Fort Collins, CO).

Laboratory Measurements

A portion of each ground sample was used for determination of laboratory concentrations of N, NDF, and ADF. NDF and ADF were determined according to the protocol outlined on Ankom Technology's (Fairport, NY) Web site (http://www.ankom.com/faqs/index.html). Nitrogen content was determined using an automated total combustion instrument (Leco, St. Joseph, MN).

Another portion of each ground sample was packed into a circular sample cup with a 2.5-cm glass cover, and scanned with a bench-top NIRS system (Model 6500, FOSS-NIRSystems, Silver Spring, MD) from 400 to 2 500 nm, but only the data in the 400–1 100-nm region were used in this study because this region closely matched that measured by the radiometer. During scanning, computer software automatically measures reflectance from an internal reference standard and then calculates, according to Equation 1, the absorbance spectra of the sample.

Equation Development/Cross-Validation and Testing

Of 107 samples in the data set, 77 were randomly selected to develop/cross-validate the calibration equations, while the remaining 30 samples were used as a test data set to validate the equations.

Development and validation of equations was performed separately for the radiometer and NIRS approaches using WINISI software (Infrasoft, 1999). The spectra from each instrument were subjected to identical mathematical treatments to facilitate direct comparison between the bench-top NIRS and radiometer approaches. Modified partial least-squares regression (Goedhart 1990; Shenk and Westerhaus 1991) was used, but was preceded by standard normal variate (SNV) scatter correction, detrending, and a 1,4,4,1 math treatment (first derivative, gap over which the derivative is calculated, number

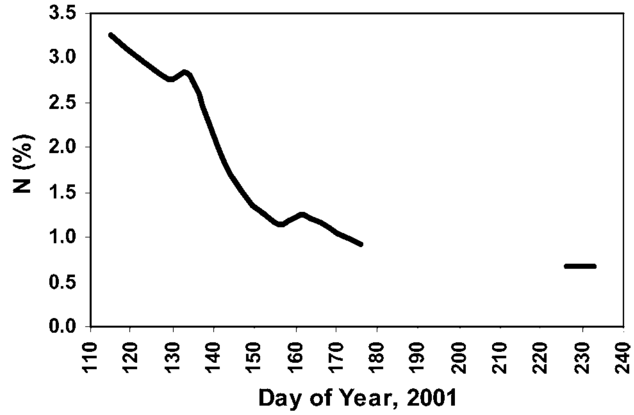

Figure 2. Time series of average weekly percent N observed during the 2001 study period. Average values from 1999 and 2000 fell within the range observed for 2001.

of points used in the first smoothing, and no second smoothing). The SNV treatment scales each spectrum so that it has a standard deviation of 1.0, which helps reduce particle size effects (Barnes et al 1989; Baker and Barnes 1990). This option was chosen because spectra from the radiometer were obtained from a canopy, whereas the spectra from the NIRS were derived from finely ground samples. Detrending helps remove linear and quadratic curvature of each spectrum (Barnes et al 1989).

Statistics reported for the equation development/cross-validation phase include the standard error of the calibration (SEC), r^2, standard error of the cross validation (SECV), and variance ratio remainder (1-VR). Statistics reported for the calibration equation test data set include the mean ($\bar{x}$), standard deviation (s), standard error of the prediction (SEP), r^2, slope, and bias.

Results

Forage Characteristics

In the 2001 study period, average weekly biomass values ranged from about 500 kg ha^{-1} (dry matter basis) on DOY 115 to about 8 000 kg ha^{-1} on DOY 176 (Fig. 1). Correspondingly, forage height ranged from a minimum of 9 to 47 cm on these days (Fig. 1). Biomass and forage height values

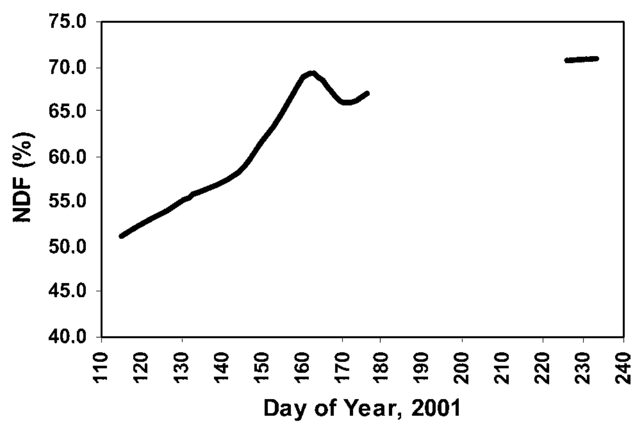

Figure 3. Time series of average weekly percent neutral detergent fiber observed during the 2001 study period. Average values from 1999 and 2000 fell within the range observed for 2001.

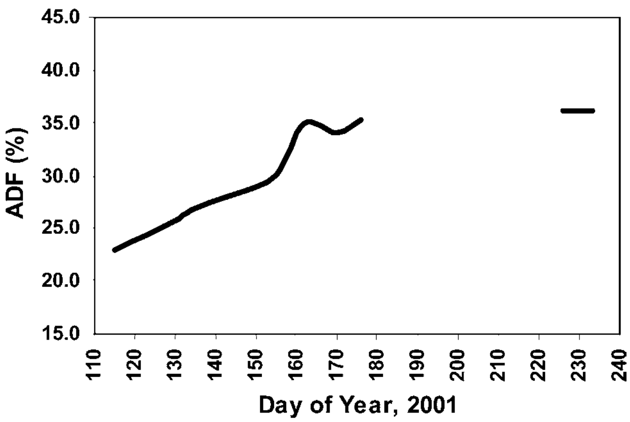

Figure 4. Time series of average weekly percent acid detergent fiber observed during the 2001 study period. Average values from 1999 and 2000 fell within the range observed for 2001.

from the 1999 and 2000 study periods fell within the range observed for the 2001 data set. Average weekly values of %N for the 2001 study period (Fig. 2) ranged from a high of 3.3 on DOY 115 to a low of about 0.7 on DOY 233. The 2001 data set shows %N decreasing at a rate of about 0.05% day^{-1} over the first 39 days of the study period (DOY 115 to 155), then decreasing at a slower rate (0.01% day^{-1}) over the remainder of the study period. Average weekly NDF increased from about 50% to 70% over the 2001 study period (Fig. 3), while ADF increased from about 23% to 36% (Fig. 4). Values of N, NDF, and ADF from the 1999 and 2000 data sets fell within the ranges observed for the 2001 data set.

Calibration/Cross-Validation Data Set

Calibration and cross-validation statistics (Table 2) indicate that the spectral data from the bench-top NIRS produced estimates of N, NDF, and ADF in closer agreement with the laboratory reference values than estimates generated using the radiometer spectral data. The radiometer SECs were higher than those observed for the NIRS for all 3 forage variables. The radiometer SEC for N was about 50% higher than that of the NIRS, but for NDF and ADF was within 10% of the SECs from the NIRS. The radiometer r^2s were all lower than their NIRS counterparts, but the spectral information from the radiometer approach accounted for 77% to 85% of the variability in the laboratory reference values, depending upon the forage variable.

Table 2. Statistics from the calibration equation development and cross-validation phase for the near-infrared spectroscopy (NIRS) and radiometer data sets ($n = 77$).

System	SEC[1] %	r^2	SEC(V) %	1-VR
		N		
NIRS	0.14	0.96	0.24	0.88
Radiometer	0.30	0.82	0.42	0.64
		NDF		
NIRS	2.15	0.84	2.63	0.77
Radiometer	2.36	0.77	3.01	0.62
		ADF		
NIRS	1.21	0.90	1.76	0.80
Radiometer	1.34	0.85	2.14	0.60

[1]SEC indicates standard error of the calibration; SEC(V), standard error of the cross-validation; 1-VR, variance ratio remainder; NDF, neutral detergent fiber; ADF, acid detergent fiber.

Table 3. Statistical results of near-infrared spectroscopy (NIRS) and radiometer calibration equations applied to the test data set ($n = 30$); mean and standard deviations are also presented for laboratory reference values as well as for NIRS and radiometer approaches.

System	$\bar{x}$	s	SEP[1]	bias
		%		
		N		
Laboratory	1.5	0.62		
NIRS	1.5	0.66	0.27	0.03
Radiometer	1.6	0.61	0.32	−0.06
		NDF		
Laboratory	65.2	4.38		
NIRS	65.1	4.17	2.40	−0.28
Radiometer	64.8	3.93	2.73	0.41
		ADF		
Laboratory	32.9	3.66		
NIRS	33.3	3.42	1.69	−0.39
Radiometer	33.1	3.09	2.06	−0.19

[1]SEP indicates standard error of the prediction; ADF, acid detergent fiber; NDF, neutral detergent fiber.

Cross-validation statistics of SECV and 1-VR were customarily higher and lower, respectively, than their SEC and r^2 counterparts (Table 2). The largest relative differences between SEC and SECV were observed for the NIRS N (about 40%) and radiometer ADF (about 37%). From the 1-VR statistic, it is observed that the NIRS approach accounted for about 15% to 24% more of the variation in the laboratory data than the radiometer approach. However, the radiometer approach accounted for 60% to 64% of the variation in the laboratory reference data.

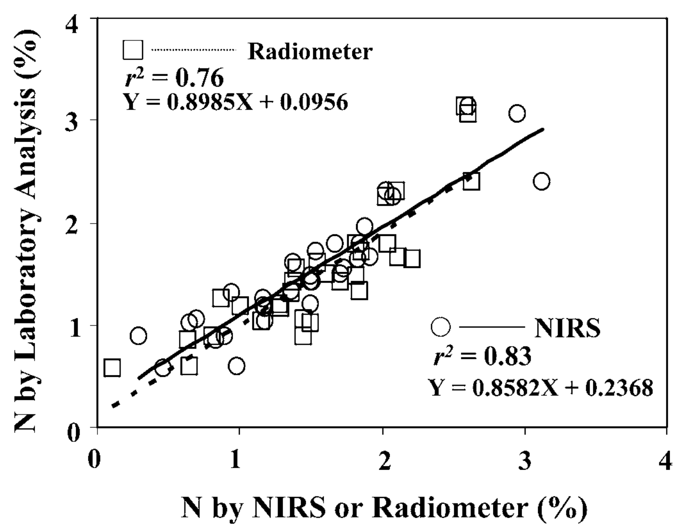

Figure 5. Scatterplot of predicted percent N values from the near-infrared spectroscopy or radiometer vs that measured in the laboratory ($n = 30$).

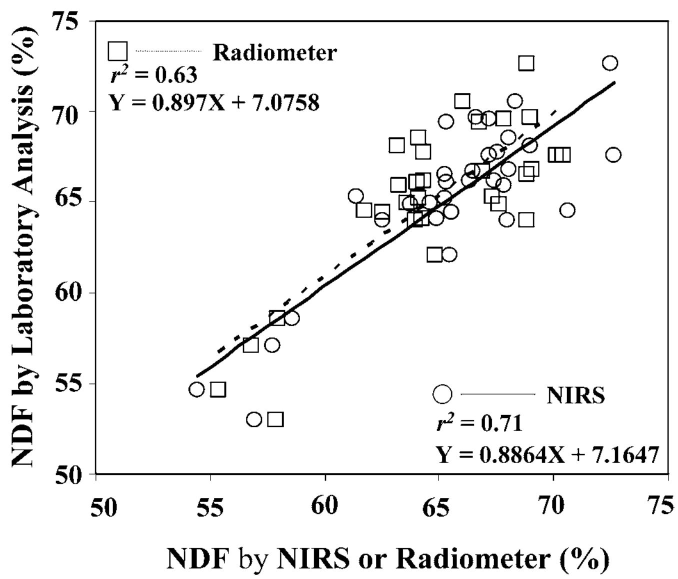

Figure 6. Scatterplot of predicted percent neutral detergent fiber values from the near-infrared spectroscopy or radiometer vs that measured in the laboratory ($n = 30$).

Test Data Set

Predicted forage composition values derived from application of the NIRS and radiometer calibration equations to the test data sets were statistically compared to laboratory reference values. It is observed (Table 3) that mean values and standard deviations derived from the NIRS and radiometer approaches are comparable to the laboratory reference values (ie, low bias). SEPs from the radiometer were from 13% to 16% larger than that of the NIRS. However, Bartlett's test showed that there was no statistical difference between the NIRS and radiometer SEPs ($0.12 \geq P \leq 0.5$). Regression analysis revealed that the NIRS r^2 values were all larger than their counterparts derived from the radiometer data (Figs. 5–7), and that both the NIRS and radiometer r^2 values from the test data set were lower than those observed in the calibration data set (Table 2). Nevertheless, the radiometer spectral data accounted for 63% to 76% of the variability in the laboratory reference data. Covariance analysis indicated that, for a given forage variable, there was no statistical difference between the NIRS and radiometer slopes ($P \geq 0.72$) and that there was no need to fit an intercept term in the regressions. Thus, the general linear models relating the NIRS and radiometer predictions to laboratory reference values were rerun forcing the regressions through the origin. This reanalysis resulted in slope values much nearer one (Table 4) for both approaches.

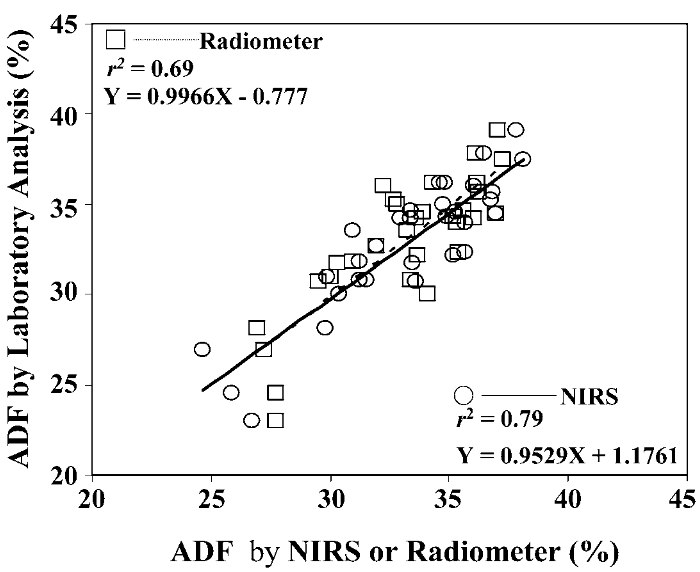

Figure 7. Scatterplot of predicted percent acid detergent fiber values from the near-infrared spectroscopy or radiometer vs that measured in the laboratory ($n = 30$).

Table 4. Slope terms after fitting regression equations with a zero-intercept.

System	Slope
N	
NIRS	0.99
Radiometer	0.952
NDF	
NIRS	0.995
Radiometer	1.006
ADF	
NIRS	0.998
Radiometer	0.994

Discussion

For this study, it was shown that calibration equations could be developed from reflectance data collected from live standing Bermuda grass canopies to predict N, NDF, and ADF. Moreover, it was observed that these predictions were comparable to those made using laboratory NIRS analysis of clipped, dried, and ground forage samples.

Although the study findings indicated that the remote sensing approach accounted for most of the variability in the laboratory reference values, less precision could be expected from this approach for at least 2 reasons. First, the sample of ground forage scanned by the NIRS instrument is very similar to that used to conduct reference chemistry for percent N, NDF, and ADF, whereas the radiometer approach measures reflectance of the forage before it is harvested. Sampling could be a significant factor in the difference between the precision of the 2 approaches. Second, the NIRS approach measures reflectance under controlled lighting conditions, but the radiometer approach measures reflectance from a live canopy under variable lighting conditions. Canopy characteristics, especially those with diverse plant populations, could contribute to variability as well. It should also be noted that, under the experimental conditions in this study, the Bermuda grass canopies were closed, minimizing any effects due to soil reflectances. In more open canopies, soil effects will have to be taken into account.

Findings from the study suggest that, for Bermuda grass, accurate remotely sensed estimates of forage composition can be obtained on standing forage from hyperspectral reflectance data. Additional warm and cool season forages will need to be studied to demonstrate the general utility of the remote sensing approach. Further testing to determine the applicability of this approach to predicting digestibility and intake is needed. If successful, this approach would allow landscape-scale nutritional information to be coupled with estimates of forage quantity, providing timely forage status assessments to the rancher, animal manager, and hay producer. Ultimately, this technology would improve profit margins by enabling timely decisions by producers of forage and animal resources. Currently, studies are under way evaluating the efficacy of this remote sensing approach to make decisions regarding the need for and timing of supplemental feeds in a Bermuda grass-based grazing system in Oklahoma.

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